

SEQUENCE LISTING

<110> Jones, David HA
Bout, Abraham

<120> Efficient Production of IgA in Recombinant Mammalian Cells

<130> 2578-6077

<150> US 09/549,463

<151> 2000-04-14

<150> US 60/129,452

<151> 1999-04-15

<160> 8

<170> PatentIn version 3.2

<210> 1

<211> 2022

<212> DNA

<213> Artificial

<220>

<223> Genomic DNA encoding heavy chain of anti-EpCAM IgA

<220>

<221> misc_feature

<222> (1)..(3)

<223> Start codon

<220>

<221> misc_feature

<222> (2020)..(2022)

<223> Stop codon

<400> 1

atggcatgcc ctggttctct gtgggcactt gtgatctcca cctgtcttga attttccatg 60

gccaggtgc agctggtgca gtctggggct gaggtgaaga agcctgggtc ctcggtgagg 120

gtctcctgca aggttcttgg aggcaccttc agcagctatg ctatcagctg ggtgcgacag 180

gcccctggac aagggttga gtggatggga gggatcatcc ctatcttgg tacagcaaac 240
 tacgcacaga agttccaggg cagagtcacg attaccgagg acgaatccac gagcacagcc 300
 tacatggagc tgagcagcct gagatctgag gacacggctg tgtattactg tgcaagagac 360
 ccgtttcttc actattgggg ccaaggtacc ctggtcaccg tctcgacagg tgagtgcggc 420
 cgtctgtgc tgggttctc cagtatagag gagaggcagg cacagactgt cctctgggg 480
 acatggcatg agggccgct cctcacagtg cattctgtgt tccagcatcc ccgaccagcc 540
 ccaaggtctt cccgtgagc ctctgcagca cccagccaga tgggaacgtg gtcatgcct 600
 gcctgtcca gggttcttc cccaggagc cactcagtgt gacctggagc gaaagcggac 660
 agggcgtgac cgccagaaac ttccacca gccaggatgc ctccggggac ctgtacacca 720
 cgagcagcca gctgaccctg ccggccacac agtccttagc cggcaagtc gtgacatgcc 780
 acgtgaagca ctacacgaat cccagccagg atgtgactgt gccctgcca ggtcagaggg 840
 caggctgggg agtggggcgg ggccaccccg tcgtgccctg acactgcgcc tgcaccctg 900
 ttccccacag ggagccgccc cttactcac accagagtgg accgcgggcc gagccccagg 960
 aggtggtggt ggacaggcca ggagggggcga ggcgggggca tggggaagca tgtgtgacc 1020
 agctcagcc atctctccac tccagttccc tcaactccac ctaccccatc tccctcaact 1080
 ccacctacc catctccctc atgtgccac cccgactgt cactgcaccg accggccctc 1140
 gaggacctgc tcttaggttc agaagcgaac ctacgtgca cactgaccgg cctgagagat 1200
 gcctcaggtg tcacctcac ctggacgccc tcaagtggga agagcgctgt tcaaggacca 1260
 cctgaccgtg acctctgtgg ctgtacagc gtgtccagt tctgtcggg ctgtgccgag 1320
 ccatggaacc atgggaagac cttacttgc actgtgcct acccgagtc caagaccccg 1380
 ctaaccgcca ccctctcaaa atccggtggg tccagaccct gctcggggcc ctgtcagt 1440
 ctctggttg caaagcatat tctggcctg cctctccct ccaatcctg ggctccagt 1500
 ctcatgcaa gtacagaggg aaactgaggc aggtgaggg gccaggacac agcccggggt 1560

gccaccaga gcagaggggc tctctcatcc cctgccagc ccctgacct ggctctctac 1620
 cctccaggaa acacattccg gcccgaggtc cacctgctgc cgccgccgtc ggaggagctg 1680
 gccctgaacg agctgggtgac gctgacgtgc ctggcacgtg gcttcagccc caaggatgtg 1740
 ctggttcgtt ggctgcaggg gtcacaggag ctgccccgcg agaagtacct gacttgggca 1800
 tcccggcagg agcccagcca gggcaccacc accttcgtg tgaccagcat actgcgcgtg 1860
 gcagccgagg actggaagaa gggggacacc ttctctgca tgggtggcca cgaggccctg 1920
 ccgtggcct tcacacagaa gaccatcgac cgcttggcgg gtaaaccac ccatgtcaat 1980
 gtgtctgttgc tcatggcgga ggtggacggc acctgctact ga 2022

<210> 2
 <211> 922
 <212> DNA
 <213> Artificial

<220>
 <223> Genomic DNA encoding light chain of anti-EpCAM IgA

<220>
 <221> misc_feature
 <222> (1)..(3)
 <223> Start Codon

<220>
 <221> misc_feature
 <222> (920)..(922)
 <223> Stop Codon

<400> 2
 atggcatgcc ctggcttctt gtgggcactt gtgatctcca cctgtcttga atttccatg 60
 gctgaaattg agctcaactca gtctccactc tccctgcccg tcacccttgg agagccggcc 120
 tccatctcct gcaggtctag tcagagcctc ctgcatagta atggatacaa ctatttgat 180
 tggtaacctgc agaagccagg gcagtctcca cagctcctga tctatttggg ttctaatecg 240

gcctccgggg tcctgacag gttcagtggc agtggatcag gcacagattt tacactgaaa 300
 atcagcagag tggaggctga ggatgttggg gtttattact gcatgcaagc tctacaaact 360
 ttactttcg gccctgggac caaggtggag atcaaacgta agtgcacttt gcggccgcta 420
 ggaagaaact caaaacatca agattttaaa tacgtttctt ggtctccttg ctataattat 480
 ctgggataag catgctgttt tctgtctgtc cctaacatgc cctgtgatta tccgcaaaca 540
 acacacccaa gggcagaact ttgttactta aacaccatcc tgtttgcttc ttcctcagg 600
 aactgtggct gcaccatctg tcttcatctt cccgccatct gatgagcagt tgaaatctgg 660
 aactgcctct gttgtgtgcc tgctgaataa cttctatccc agagaggcca agtacagtg 720
 gaaggtggat aacgccctcc aatcgggtaa ctcccaggag agtgtcacag agcaggacag 780
 caaggacagc acctacagcc tcagcagcac cctgacgctg agcaaagcag actacgagaa 840
 acacaaagtc tacgctgcg aagtcacca tcagggcctg agctcgcccg tcacaaagag 900
 cttcaacagg ggagagtgtt ag 922

<210> 3
 <211> 489
 <212> PRT
 <213> artificial

<220>
 <223> Amino acid sequence anti-EpCAM IgA heavy chain

<220>
 <221> MISC_FEATURE
 <222> (1)..(21)
 <223> leader peptide

<220>
 <221> MISC_FEATURE
 <222> (22)..(136)
 <223> VH Region

<220>

<221> MISC_FEATURE

<222> (137)..(238)

<223> CH1 Region

<220>

<221> MISC_FEATURE

<222> (239)..(359)

<223> CH2 Region

<220>

<221> MISC_FEATURE

<222> (360)..(489)

<223> CH3 Region

<400> 3

Met Ala Cys Pro Gly Phe Leu Trp Ala Leu Val Ile Ser Thr Cys Leu
1 5 10 15

Glu Phe Ser Met Ala Gln Val Gln Leu Val Gln Ser Gly Ala Glu Val
20 25 30

Lys Lys Pro Gly Ser Ser Val Arg Val Ser Cys Lys Ala Ser Gly Gly
35 40 45

Thr Phe Ser Ser Tyr Ala Ile Ser Trp Val Arg Gln Ala Pro Gly Gln
50 55 60

Gly Leu Glu Trp Met Gly Gly Ile Ile Pro Ile Phe Gly Thr Ala Asn
65 70 75 80

Tyr Ala Gln Lys Phe Gln Gly Arg Val Thr Ile Thr Ala Asp Glu Ser
85 90 95

Thr Ser Thr Ala Tyr Met Glu Leu Ser Ser Leu Arg Ser Glu Asp Thr
100 105 110

Ala Val Tyr Tyr Cys Ala Arg Asp Pro Phe Leu His Tyr Trp Gly Gln
115 120 125

Gly Thr Leu Val Thr Val Ser Thr Ala Ser Pro Thr Ser Pro Lys Val
130 135 140

Phe Pro Leu Ser Leu Cys Ser Thr Gln Pro Asp Gly Asn Val Val Ile

145 150 155 160
 Ala Cys Leu Val Gln Gly Phe Phe Pro Gln Glu Pro Leu Ser Val Thr
 165 170 175
 Trp Ser Glu Ser Gly Gln Gly Val Thr Ala Arg Asn Phe Pro Pro Ser
 180 185 190
 Gln Asp Ala Ser Gly Asp Leu Tyr Thr Thr Ser Ser Gln Leu Thr Leu
 195 200 205
 Pro Ala Thr Gln Cys Leu Ala Gly Lys Ser Val Thr Cys His Val Lys
 210 215 220
 His Tyr Thr Asn Pro Ser Gln Asp Val Thr Val Pro Cys Pro Val Pro
 225 230 235 240
 Ser Thr Pro Pro Thr Pro Ser Pro Ser Thr Pro Pro Thr Pro Ser Pro
 245 250 255
 Ser Cys Cys His Pro Arg Leu Ser Leu His Arg Pro Ala Leu Glu Asp
 260 265 270
 Leu Leu Leu Gly Ser Glu Ala Asn Leu Thr Cys Thr Leu Thr Gly Leu
 275 280 285
 Arg Asp Ala Ser Gly Val Thr Phe Thr Trp Thr Pro Ser Ser Gly Lys
 290 295 300
 Ser Ala Val Gln Gly Pro Pro Asp Arg Asp Leu Cys Gly Cys Tyr Ser
 305 310 315 320
 Val Ser Ser Val Leu Ser Gly Cys Ala Glu Pro Trp Asn His Gly Lys
 325 330 335
 Thr Phe Thr Cys Thr Ala Ala Tyr Pro Glu Ser Lys Thr Pro Leu Thr
 340 345 350
 Ala Thr Leu Ser Lys Ser Gly Asn Thr Phe Arg Pro Glu Val His Leu
 355 360 365
 Leu Pro Pro Pro Ser Glu Glu Leu Ala Leu Asn Glu Leu Val Thr Leu
 370 375 380

Thr Cys Leu Ala Arg Gly Phe Ser Pro Lys Asp Val Leu Val Arg Trp
385 390 395 400

Leu Gln Gly Ser Gln Glu Leu Pro Arg Glu Lys Tyr Leu Thr Trp Ala
 405 410 415

Ser Arg Gln Glu Pro Ser Gln Gly Thr Thr Thr Phe Ala Val Thr Ser
 420 425 430

Ile Leu Arg Val Ala Ala Glu Asp Trp Lys Lys Gly Asp Thr Phe Ser
 435 440 445

Cys Met Val Gly His Glu Ala Leu Pro Leu Ala Phe Thr Gln Lys Thr
 450 455 460

Ile Asp Arg Leu Ala Gly Lys Pro Thr His Val Asn Val Ser Val Val
465 470 475 480

Met Ala Glu Val Asp Gly Thr Cys Tyr
 485

<210> 4

<211> 239

<212> PRT

<213> Artificial

<220>

<223> amino acid sequence anti-EpCAM IgA light chain

<220>

<221> MISC_FEATURE

<222> (1)..(21)

<223> leader peptide

<220>

<221> MISC_FEATURE

<222> (22)..(132)

<223> VL region

<220>

<221> MISC_FEATURE

<222> (133)..(239)

<223> CL region

<400> 4

Met Ala Cys Pro Gly Phe Leu Trp Ala Leu Val Ile Ser Thr Cys Leu
1 5 10 15

Glu Phe Ser Met Ala Glu Ile Glu Leu Thr Gln Ser Pro Leu Ser Leu
20 25 30

Pro Val Thr Pro Gly Glu Pro Ala Ser Ile Ser Cys Arg Ser Ser Gln
35 40 45

Ser Leu Leu His Ser Asn Gly Tyr Asn Tyr Leu Asp Trp Tyr Leu Gln
50 55 60

Lys Pro Gly Gln Ser Pro Gln Leu Leu Ile Tyr Leu Gly Ser Asn Arg
65 70 75 80

Ala Ser Gly Val Pro Asp Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp
85 90 95

Phe Thr Leu Lys Ile Ser Arg Val Glu Ala Glu Asp Val Gly Val Tyr
100 105 110

Tyr Cys Met Gln Ala Leu Gln Thr Phe Thr Phe Gly Pro Gly Thr Lys
115 120 125

Val Glu Ile Lys Arg Thr Val Ala Ala Pro Ser Val Phe Ile Phe Pro
130 135 140

Pro Ser Asp Glu Gln Leu Lys Ser Gly Thr Ala Ser Val Val Cys Leu
145 150 155 160

Leu Asn Asn Phe Tyr Pro Arg Glu Ala Lys Val Gln Trp Lys Val Asp
165 170 175

Asn Ala Leu Gln Ser Gly Asn Ser Gln Glu Ser Val Thr Glu Gln Asp
180 185 190

Ser Lys Asp Ser Thr Tyr Ser Leu Ser Ser Thr Leu Thr Leu Ser Lys
195 200 205

Ala Asp Tyr Glu Lys His Lys Val Tyr Ala Cys Glu Val Thr His Gln
210 215 220

Gly Leu Ser Ser Pro Val Thr Lys Ser Phe Asn Arg Gly Glu Cys
225 230 235

<210> 5
<211> 38
<212> DNA
<213> Artificial

<220>

<223> E001 forward primer for amplification of light chain

<400> 5
cctggcgcgc caccatggca tgccctggct tcctgtgg 38

<210> 6
<211> 32
<212> DNA
<213> Artificial

<220>

<223> E002 reverse primer for amplification of light chain

<400> 6
ccgggttaac taacactctc ccctgttgaa gc 32

<210> 7
<211> 39
<212> DNA
<213> Artificial

<220>

<223> E003 forward primer for amplification of heavy chain

<400> 7

ggaggatccg ccaccatggc atgcctggc ttcctgtgg

39

<210> 8

<211> 29

<212> DNA

<213> Artificial

<220>

<223> P01 reverse primer for amplification of heavy chain

<400> 8

ggaccgctag ctca tagca ggtgccgac

29